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EXAMINER	
AKHAVAN, RAMIN	
ART UNIT	**PAPER NUMBER**
1636	

DATE MAILED: 08/24/2004

Please find below and/or attached an Office communication concerning this application or proceeding.

Applicant 08/23/04 RX

Office Action Summary	Application No.	Applicant(s)	
	10/037,243	FREIMUTH ET AL.	
	Examiner	Art Unit	
	Ramin (Ray) Akhavan	1636	

-- *The MAILING DATE of this communication appears on the cover sheet with the correspondence address* --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *02 August 2004*.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *53-64* is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *53-64* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☒ The specification is objected to by the Examiner.

10)☒ The drawing(s) filed on *03 June 2002* is/are: a)☐ accepted or b)☒ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☒ Information Disclosure Statement(s) (PTO-1449 or PTO/SB/08) Paper No(s)/Mail Date *06/14 and 7/1/02*.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____ .
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☐ Other: _____.

DETAILED ACTION

Acknowledgment is made of amendments, filed 06/03/2002, inserting sequence identifiers in the claims and the specification.

Election/Restrictions

Applicant's election without traverse of Group 5, claims 53-64, in the reply filed on 08/02/2004, is acknowledged, including election of species of the peptide extension consisting of SEQ ID NO: 6, with respect to claim 64. Claims 53-64 are pending and under consideration in this action. Claims 1-52 and 65-86 are withdrawn as drawn to non-elected subject matter.

Drawings

The drawings are objected to because the actual drawings do not include the appropriate references (i.e. "Fig. 1", "Fig. 2" or "Fig. 3"). The corrected drawings should include the appropriate reference. Corrected drawing sheets in compliance with 37 CFR 1.121(d) are required in reply to the Office action to avoid abandonment of the application. Any amended replacement drawing should include all of the figures appearing on the immediate prior version of the sheet, even if only one figure is being amended. The figure or figure number of an amended drawing should not be labeled as "amended." If a drawing figure is to be canceled, the appropriate figure must be removed from the replacement sheet, and where necessary, the remaining figures must be renumbered and appropriate changes made to the brief description of the several views of the drawings for consistency. Additional replacement sheets may be

necessary to show the renumbering of the remaining figures. The replacement sheet(s) should be labeled "Replacement Sheet" in the page header (as per 37 CFR 1.84(c)) so as not to obstruct any portion of the drawing figures. If the changes are not accepted by the examiner, the applicant will be notified and informed of any required corrective action in the next Office action. The objection to the drawings will not be held in abeyance.

Specification

The abstract of the disclosure is objected to because it exceeds the maximum allowed 150 words. Correction is required. See MPEP § 608.01(b).

The disclosure is objected to because of the following informalities: The disclosure, on page 46, contains an embedded hyperlink and/or other form of browser-executable code. Applicant is required to delete the embedded hyperlink and/or other form of browser-executable code. See MPEP § 608.01. Appropriate correction is required.

Claim Rejections - 35 USC § 112

The following is a quotation of the second paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter, which the applicant regards as his invention.

1. **Claims 53-64 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.**

Base claim 53 recites the limitation "said peptide extension"; where the expression vector comprises a cloning site for inserting in-frame another nucleic acid of interest.

There is insufficient antecedent basis for this limitation in the claim, because the preamble recites "a nucleic acid sequence encoding a peptide extension" which is distinguishable from the *encoded* peptide extension. As such the nucleic acid of interest would not actually be in-frame with a peptide extension but with the nucleic acid encoding the peptide extension. It would be remedial to include the term, "nucleic acid encoding" before "said peptide extension", so as to maintain proper antecedent support.

Furthermore, claim 53 recites the phrase, "the peptide extension having a net negative charge ranging from –2 to –20", which confers ambiguity and vagueness. First, it is unclear whether the peptide extension is of a particular size. For example, can the extension be a 100mer or a 500mer? Second, it is unclear under what conditions (e.g. pH ranges, temperature, salt) the peptide extension has a net negative charge in the prescribed range. This is relevant as expression at different pH or temperature, for example, could affect protein solubility. Third, it is unclear if the polypeptide of interest is can be of any size. For example, beyond a certain limit there would be concerns with respect to the vector actually being able to accommodate the gene encoding the polypeptide of interest. In sum, it is unclear how the phrase is to be interpreted in determining the claims' metes and bounds.

In addition, claim 62 recites the phrase, "activity promoting portions thereof" when referring to the 57 residue of T7 gene 10B protein. It is unclear how the term, "activity" is to be interpreted in determining the claim's metes and bounds. There does not appear to be any direct reference to the "activity" to which the claim is directed.

Additionally, claim 63 recites the phrase, "active portions thereof", which does not appear to be particularly defined in the specification. Again, it is unclear to what "activity" the

claim is delimited, therefore, as written the claim is vague and indefinite. There is reference in the specification to a proteins' biological activity, but this description does not apply to the peptide extensions. (e.g. Spec. p. 12, ¶ 2).

The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

2. Claims 62-64 are rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the written description requirement.

The claims contain subject matter, which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventors, at the time the application was filed, had possession of the claimed invention. More particularly, the claims are directed to a genus of peptide extensions having a net negative charge (-2 to –20), nucleic acid structures comprising portions of the T7 gene 10B protein having some "activity". This is a genus claim in terms of *any* portion of the 57 residue of the 10B protein having *any* activity. The specification does not contain any examples of such structures having *any* activity. Thus the disclosure is not descriptive of the complete structure of a representative number of species, which the claims encompass, as one of ordinary skill in the art cannot envision all the variants that are possible as "portions" from the 57 residue template capable of an activity, based on the teachings in the specification. The written description requirement for a claimed genus may be satisfied by sufficient description of a representative number of species by actual reduction to practice, reduction to drawings or by disclosure relevant identifying characteristics, i.e. structure

or other physical and/or chemical properties, by functional characteristics coupled with a known or disclosed correlation between function and structure or by a combination of such identifying characteristics sufficient to show applicant was in possession of the claimed genus.

The only activity disclosed in the specification is that of enhancing protein solubility within a bacterial host. Given the enormous breadth of the structures/activities encompassed by the rejected claims, and given the limited description from the instant specification of such structures as correlated to having any activity, the skilled artisan would not have been able to envision a sufficient number of specific embodiments to describe the broadly claimed genus. Moreover, an applicant claiming a biotechnological invention cannot necessarily claim a genus after only describing a limited number of species because there may be unpredictability in the results obtained from other species. Therefore, the skilled artisan would reasonably have concluded that applicants were not in possession of the claimed invention.

3. **Claims 53-64 are rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for disclosed peptide extensions promoting protein solubility under neutral pH conditions in *E. coli*, does not reasonably provide enablement for any peptide extension, having any size, expressed in any host, linked to any size target protein, expressed under any conditions and having any activity.**

The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the invention commensurate in scope with these claims. The test for enablement is whether one skilled in the art could make and use the claimed invention from the disclosure in the specification coupled with information known in

the art without undue experimentation. *United States v Telectronics Inc.*, 8 USPQ2d 1217 (Fed. Cir. 1988). Whether undue experimentation is required is not based upon a single factor but instead is a conclusion reached by weighing many factors which are outlined in *Ex parte Forman*, 230 USPQ 546 (Bd. Pat. App. & Inter. 1986) and *In re Wands*, 8 USPQ2d 1400 (Fed. Cir. 1988). The factors include the following:

Scope/Breadth of the claims. The claims' scope is both wide and deep. The claims are directed to expression vectors comprising any peptide extension having the prescribed range of net negative charge, where the peptide extensions can be any size, linked to a protein of interest of any size, where the fusion proteins are expressed in any host, under any conditions (e.g. pH or temperature) and the extensions can have any "activity".

Nature of the invention. The invention is directed protein expression vectors. The only disclosed utility for the vector compositions is directed to properly folding and enhanced solubility based on fusion protein technology, particularly to preclude inclusion body formation, incorporating a peptide extension that inheres a net negative charge. In essence, the peptide extension can be thought of as a "solubilizing partner".

State of the art/Unpredictability of the art. The state of the art of protein production is somewhat developed in regard to particular host organisms, e.g. *E. coli*. Indeed, even with regard to *E. coli* based protein expression, many fundamental aspects of physiology remain to be uncovered and will affect progress in optimizing this bacterium for protein expression. (e.g. Baneyx, F. Curr. Opin. Biotech. 1999; 10:411-421, p. 418, col. 2). However, there are vagaries as to protein production that often require considerable, costly and lengthy experimentation, each

of which would necessarily be exacerbated for host organisms for which there is little knowledge in the art with respect to recombinant protein production.

Attaching any sized peptide extension, fused to a target protein of any size would not necessarily result in expression or enhanced folding/solubility in any host organisms. For example, even within well characterized systems, such as in *E. coli*, expression of larger proteins in a cell is often inefficient or unsuccessful, particularly as the protein of interest or expressed protein in general increases in size or is smaller but a multi-domain protein. (e.g. Frydman, J. Annu. Rev. Biochem. 2001; 70:603-47; p. 621, ¶ 2; present in IDS, thus a copy is not being submitted herewith). Furthermore, depending on the size of the target gene encoding the target protein, a vector may not even be available to effectuate protein expression. In the same vein, the vector used may become unstable in a particular cell type, or the encoded protein may be toxic to a particular host, resulting in vector loss, thus no protein expression. (e.g. Baneyx, F. Curr. Opin. Biotech. 1999; 10:411-421, p. 411, col. 2). Arguendo, even if such unpredictability for a particular host such as *E. coli*, could be worked out with routine experimentation, such is not necessarily the case for any cell system. Furthermore, with respect to fusion protein technology to enhance proper folding, to preclude inclusion body formation, all fusion partners for a particular target protein will not equally alleviate inclusion body formation. (Id. p. 471, ¶ 1). Ironically, increased solubility for a particular fusion protein may itself lead to product aggregation as more of the protein would accumulate intra-cellularly, which in turn can lead to inclusion body formation. (e.g. Swartz, JR. Curr. Opin. Biotech. 2001; 12:195-201, at 201, col. 2; ¶, 4). In addition, even with proper folding, the fused protein may not contain native target

protein activity, due to formation and isomerization of disulfide bonds, which itself can depend on the host cell or the size and number of protein domains.

In regard to protein expression, an area of unpredictability may be mRNA stability or eve more importantly translation initiation, which could differ, based on the size of the fusion protein (whether through variability in the extension or the target protein), or based on the type of cell species/strains being used. (See generally, Swartz, JR. Curr. Opin. Biotech. 2001, at pp. 196-7; noting mRNA secondary structure can block ribosome binding caused significant decrease in expression). Importantly, in regard to enhancing proper folding to preclude inclusion body formation, "Finding the optimal refolding conditions is still relatively empirical, with the best results obtained from evaluating a matrix of conditions affecting solubility and disulfide-bond formation and isomerization."[1] (Id. at p. 197, col. 2, last ¶). With respect to the target protein size and characteristics affecting protein folding, thus production, solubilizing partners do not always preclude inclusion body formation, thus enhance solubility. (Thomas et al. Appl. Biochem. Biotech. 1997, 66:197-238; p. 201, ¶ 2). While the "partners" studied in the art may not be the same peptide extensions as in instant application, the salient analogous point is, that while a fusion construct does aid in proper folding/solubility for one protein, it does not necessarily do so for any protein in any cell system.

Amount of guidance provided. The specification provides prophetic guidance as to expression of proteins in any host cell and indicates that any size peptide extension can be used, where the extension having has the prescribed negative charge deemed necessary to provide enhanced solubility/folding. (e.g. Spec. p. 18, ¶ 2). The specification does not provide any

guidance as to what would be the size limitations for the peptide extensions, target proteins relative to particular vectors (i.e. vector loss/maintenance) or other cells whether eukaryotes or prokaryotes. Nor does the specification contemplate differentials in regard to temperature or pH, for example, in different cellular hosts, in effecting the charge characteristics for peptide extensions.

Number of working examples. The specification provides three peptide extensions (i.e. T7A, T7B and T7C, or SEQ ID NOs: 6 and 5), which are shown to enhance solubility of the a single target protein – CAR D1 – where protein expression is induced in *E. coli* cells, at 25° and 37° C. (Spec. pp. 27-29; showing expression of CAR D1-T7B and -T7C fusion proteins).

Amount of Experimentation Required. The level of skill in the art required to practice the claimed invention is high. Given the unsolved hurdles to successful practicing of the invention, the level of unpredictability in the art and lack of working examples commensurate with the scope of the claims, it must be considered that the skilled artisan would be required to conduct trial and error experimentation of an undue nature in order to attempt to practice the claimed invention commensurate with the scope of the claims.

Claim Rejections - 35 USC § 102

The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

> A person shall be entitled to a patent unless –
>
> (b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

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[1] This quote is only being proffered to show unpredictability in the art, not that Applicant is required to exemplify the *best* conditions for practicing the invention.

4. Claims 53-61 are rejected under 35 U.S.C. 102(b) as being anticipated by Rechsteiner et al. (US 5,366,871; see whole document; hereinafter the '871 patent).

The claims are drawn to an expression vector encoding a fusion protein comprising a peptide extension and a protein of interest. This limitation is interpreted as broadly as reasonable in that where a fusion construct is taught having two distinct amino acid sequences in-frame, either sequence can constitute an extension or the protein of interest (e.g. A-B fusion, A can be an extension while B is the protein of interest or vice versa). Furthermore, net charge is interpreted to mean the sum of the basic and acidic charges, i.e., one positive and one negative based on the individual amino acid residues comprising the peptide. Peptide of interest is not restricted to any particular size, thus can be a protein or polypeptide.

The '871 patent teaches an expression vector to use in a cell where a peptide extension is linked to a gene of interest (e.g. Abstract; Fig 1). More particularly, the vector is an M13 vector encoding a fusion protein of an ubiquitin peptide extension in frame with a protein of interest – cRAS. (e.g. Fig. 1; col. 8, Example 1). Expression is observed in prokaryotes (e.g. col. 9, Example 2) or it can also be in eukaryotes (e.g. col. 13, Example 10). The vector contains a polylinker or multiple cloning site. (e.g. col. 8, l. 55). Furthermore, the ubiquitin peptide extension contains the amino acid residues Ser-Glu-Glu-Glu-Glu, which would necessarily have a net negative charge of –4, as it contains four acidic or negative side chains. Moreover, it is an intrinsic property of peptides in solution that if the pH ranges in solution is altered (e.g. by adding buffer to a solution) that the net charge for the peptide is changed (e.g. more or less negative). In addition, the fusion construct comprises additional peptide extensions consisting of the amino acid sequence, Pro-Gly-Cys-Met-Ser-Cys-Lys-Cys-Val-Leu-Ser (e.g. col. 8, Example

1), as well as multiple other peptide extensions as represented by SEQ ID NOs: 1-9, each of which would inhere a different net negative charge. For example, the peptide extension represented by SEQ ID NO: 4 would contain a net negative charge of –5. (e.g. col. 17, SEQ ID NO: 4).

As noted above, an intrinsic property for any peptide sequence in a solution is that at different pH values the peptide has a different net charge. Therefore, the various peptides taught in the '871 patent could intrinsically contain a different net negative charge. Because the Office does not have the facilities for examining and comparing the applicant's product with the products of the prior art, the burden is on the applicant to show a novel or unobvious difference between the claimed products and the products of the prior art (e.g. that the peptide extensions of the prior art do not possess the same material structural and functional characteristics of the claimed product). See in re Best, 562 F.2d 1252, 195 USPQ 430 (CCPA 1977). In sum, the '871 patent anticipates the rejected claims.

5. Claims 53-54 and 56-63 are rejected under 35 U.S.C. 102(b) as being anticipated by Studier et al. (US 5,766,905; see whole document; hereinafter the '905 patent).

Additional embodiments are directed to the peptide extension comprising the c-terminus portion of the T7 10B protein. In addition, the limitation "about 61 amino acid" is interpreted as broadly as reasonable in light of the term "about", as not excluding peptide extensions exceeding 61 amino acids.

The '905 patent teaches expression vectors for expression of fusion proteins where a genes encoding a protein of interest are fused in-frame at the N-terminus of the 348 amino acid

10B protein. (e.g. col. 3, ll. 8-29). In addition, a modified version of the 10B protein (frame shift site at amino acid 341 had been removed) is taught. (e.g. col. 3, l. 13). As noted above, version of the 10B protein extension taught in the '905 patent would necessarily inhere a net charge depending on variability in pH. Thus at the appropriate pH, the 348 amino acid peptide extension can have a negative charge anywhere from –2 to –20. Because the Office does not have the facilities for examining and comparing the applicant's product with the products of the prior art, the burden is on the applicant to show a novel or unobvious difference between the claimed products and the products of the prior art (e.g. that the products of the prior art do not possess the same material structural and functional characteristics of the claimed product). See in re Best, 562 F.2d 1252, 195 USPQ 430 (CCPA 1977). In sum, the '905 patent anticipates the rejected claims.

6. Claims 53-61 rejected under 35 U.S.C. 102(b) as being anticipated by Harrison et al. (US 5,989,868; see whole document; hereinafter the '868 patent).

The '868 patent teaches a vector system for expression of fusion proteins. (e.g. Abstract; col. 2, ll. 10-63). More particularly, the '868 patent teaches a fusion construct where a protein of interest is fused to the carboxy terminus of a carrier peptide, i.e. peptide extension. (e.g. col. 9, ll. 8-15; col. 11, ll. 1). The expression can be adapted to be in either prokaryotes or eukaryotes. (e.g. col. 9, l. 25). Furthermore, the carrier proteins (peptide extensions) can be selected from a group of protein varying in size from 146 to 495 amino acid residues. (e.g. col. 4, Table 1). Thus each stretch of amino acids can necessarily have a net negative charge based on the side chains of the amino acid residues. Because the Office does not have the facilities for examining and comparing the applicant's product with the products of the prior art, the burden is on the

applicant to show a novel or unobvious difference between the claimed products and the products of the prior art (e.g. that the products of the prior art do not possess the same material structural and functional characteristics of the claimed product). See in re Best, 562 F.2d 1252, 195 USPQ 430 (CCPA 1977). In sum, the '868 patent anticipates the rejected claims.

Conclusion

No claims are allowed.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Ramin (Ray) Akhavan whose telephone number is 571-272-0766. The examiner can normally be reached on Monday- Friday from 8:00-4:30. If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Remy Yucel, Ph.D. can be reached on 571-272-0781. The fax phone number for the organization where this application or proceeding is assigned is 703-872-9306.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free).

Ray Akhavan/AU 1636

GERRY LEFFERS
PRIMARY EXAMINER